IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s):	Robert H. Harris	**Examiner:**	David Lukton
Serial No:	10/688,638	**Art Unit:**	1654
Filed:	October 17, 2003	**Docket:**	13095B
For:	NEW USES FOR AMINO ACID ANTICONVULSANTS		

Confirmation No: 2598

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION OF ROBERT H. HARRIS
UNDER 37 C.F.R. §1.132

Sir:

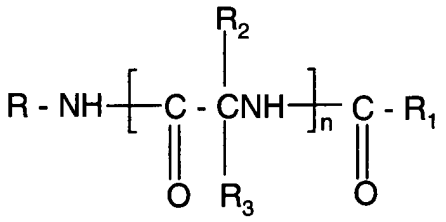
Dr. Robert H. Harris declares and says as follows:

1. I am the inventor of the subject matter of the above-identified application, and I have complete knowledge of all aspects of the invention embodied therein.

2. I have received the degree of the Doctor of Philosophy in Bio-Chemistry from Rutgers University in 1977.

3. I currently am the owner of and hold the title of President at Harris FRC Corporation.

4. The present application is directed, inter alia, to the method of treating a patient suffering from bipolar disorder comprising administering thereto a therapeutically effective amount of a compound for treating bipolar disorder, said compound having the formula:



wherein

R is unsubstituted or is substituted with at least one electron withdrawing group or electron donating group;

R_1 is lower alkyl and is unsubstituted or substituted with at least one electron withdrawing group or electron donating group;

R_2 is hydrogen, lower alkyl, lower alkenyl, lower alkynyl, aryl lower alkyl, aryl, halo, heterocyclic, heterocyclic lower alkyl, lower alkyl heterocyclic, lower cycloalkyl, lower cycloalkyl lower alkyl, or Z-Y_x

R_3 is lower alkyl, lower alkenyl, lower alkynyl, aryl, aryl lower alkyl, halo, heterocyclic, heterocyclic lower alkyl, lower alkyl heterocyclic, lower cycloalkyl, lower cycloalkyl lower alkyl or ZY; wherein R_2 and R_3 may be unsubstituted or substituted with at least one electron withdrawing group or electron donating group , and wherein heterocyclic in R_2 and R_3 is furyl, thienyl, pyrazolyl, pyrrolyl, imidazolyl, indolyl, thiazolyl, oxazolyl, isothiazolyl, isoxazolyl, piperidyl, pyrrolinyl, piperazinyl, quinolyl, triazolyl, tetrazolyl, isoquinolyl, benzofuryl, benzothienyl, morpholinyl, benzoxazolyl, tetrahydrofuryl, pyranyl, indazolyl, purinyl, indolinyl, pyrazolindinyl, imidazolinyl, imidazolindinyl, pyrrolidinyl, furazanyl, N-methylindolyl, methylfuryl, pyridazinyl, pyrimidinyl, pyrazinyl, epoxy, aziridino, oxetanyl or azetidinyl;

Z is O, S, or NR_6'

Y is hydrogen, lower alkyl, aryl, aryl lower alkyl, lower alkenyl, or lower alkynyl, and Y may be unsubstituted or substituted with an electron donating group or an electron withdrawing group, or

ZY taken together is $NR_4NR_5R_7$, NR_4OR_5, or ONR_4R_7.

R_6' is hydrogen or lower alkyl and R_6' may be unsubstituted or substituted with an electron withdrawing group or an electron donating group;

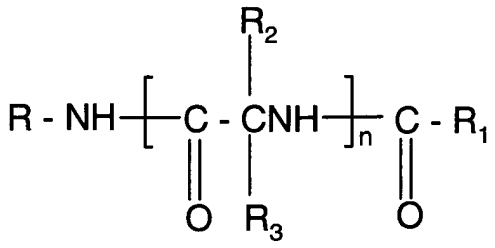
R_4 and R_5 are independently hydrogen, lower alkyl, aryl, aryl lower alkyl, lower alkenyl, or lower alkynyl, wherein R_4 and R_5 are independently unsubstituted or substituted with an electron withdrawing group or an electron donating group; and

R_7 is $COOR_8$, COR_8, hydrogen, lower alkyl, aryl, or aryl lower wherein R_7 may be unsubstituted or substituted with an electron withdrawing group or electron donating group;

R_8 is hydrogen or lower alkyl, or aryl lower alkyl, and the aryl or alkyl group may be unsubstituted or substituted with an electron withdrawing group or an electron donating group; and

n is 1; wherein the electron withdrawing group and electron donating group are selected from the group consisting of halo, nitro, lower alkenyl, lower alkynyl, formyl, aryl, trifluoromethyl, aryl lower alkanoyl, lower alkoxy carbonyl, hydroxy, lower alkoxy, lower alkyl, mercapto, lower alkylthio and lower alkyldithio.

It is also directed, inter alia, to method of treating a patient suffering from bipolar disorder comprising administering to said patient a therapeutically amount of a compound of the formula:



wherein

R is aryl lower alkyl and R is unsubstituted or is substituted with at least one electron withdrawing group or electron donating group selected from the group consisting of halo, nitro, lower alkenyl, lower alkynyl, formyl, aryl, trifluoromethyl, lower alkoxy carbonyl, aryl lower alkanoyl, hydroxy, lower alkoxy, lower alkyl, mercapto, lower alkylthio, and lower alkyldithio;

R_1 is methyl, and is unsubstituted or substituted with an electron donating group or an electron withdrawing group selected from the group consisting of halo, nitro, lower alkenyl, lower alkynyl, formyl, aryl, trifluoromethyl, lower alkoxy carbonyl, aryl lower alkanoyl, hydroxy, lower alkoxy, lower alkyl, mercapto, lower alkylthio, and lower alkyldithio;

R_2 is hydrogen, lower alkyl, lower alkenyl, lower alkynyl, aryl, aryl lower alkyl, halo, heterocyclic, heterocyclic lower alkyl, lower alkyl heterocyclic, lower cycloalkyl, lower cycloalkyl lower alkyl, or ZY;

R_3 is lower alkyl, lower alkenyl, lower alkynyl, aryl, aryl lower alkyl, halo, heterocyclic, heterocyclic lower alkyl, lower alkyl heterocyclic, lower cycloalkyl, lower cycloalkyl lower alkyl or ZY;

wherein R_2 and R_3 may be unsubstituted or substituted with at least one electron withdrawing group or electron donating group and wherein heterocyclic in R_2 and R_3 is furyl, thienyl, pyrazolyl, pyrrolyl, imidazolyl, indolyl, thiazolyl, oxazolyl, isothiazolyl, isoxazolyl, piperidyl, pyrrolinyl, piperazinyl, quinolyl, triazolyl, tetrazolyl, isoquinolyl, benzofuryl, benzothienyl, morpholinyl, benzoxazolyl, tetrahydrofuryl, pyranyl, indazolyl, purinyl, indolinyl,

pyrazolindinyl, imidazolinyl, imidazolindinyl, pyrrolidinyl, furazanyl, N-methylindolyl, methylfuryl, pyridazinyl, pyrimidinyl, pyrazinyl, epoxy, aziridino, oxetanyl or azetidinyl;

Z is O or NR_6';

Y is hydrogen, lower alkyl, aryl, aryl lower alkyl, lower alkenyl or lower alkynyl, and Y may be unsubstituted or substituted with an electron donating group or an electron withdrawing group, or

ZY taken together is $NR_4NR_5R_7$, NR_4OR_5, or ONR_4R_7;

R_6' is hydrogen or lower alkyl;

R_4 and R_5 are independently hydrogen, lower alkyl, aryl, aryl lower alkyl, lower alkenyl, or lower alkynyl, and R_4 and R_5 may be independently unsubstituted or substituted with an electron withdrawing group or an electron donating group;

R_7 is $COOR_8$, COR_8, hydrogen, lower alkyl, aryl or aryl lower alkyl, which R_7 may be unsubstituted or substituted with an electron withdrawing group or electron donating group;

R_8 is hydrogen or lower alkyl, or aryl lower alkyl, and the aryl or alkyl group may be unsubstituted or substituted with an electron withdrawing group or an electron donating group; and

n is 1.

5. I have reviewed the Official Action dated September 23, 2005. I have been requested by counsel to submit additional data and to more particularly illustrate that the compounds discussed hereinabove are useful for the treatment of bipolar disorders as described in the instant application.

6. The experiments described in the application and in the present Declaration were conducted under my direct supervision and control. I was fully cognizant of all aspects of the experiments performed and I have interpreted the data as described hereinbelow.

7. The first set of experiments utilizes a representative compound within the scope of the formula described in Paragraph 4. More specifically, (2R)-2-(acetylamino)-N-[(3-fluorophenyl)methyl]-3-methoxypropionamide, a representative compound of the present invention, which is designated as Compound 1 herein, was tested according to the following procedure.

8. The protocol is based on the assay developed by Williams, et al., *Nature*, 417: 292-295 (2002) attached hereto as Exhibit A and subsequently extended by Cheng et al., *Mol. Cell. Neurosci.*, 29: 155-161 (2005) attached hereto as Exhibit B and Di Daniel et al., *Mol. Cell. Neurosci.*, (in press) (2006), attached hereto as Exhibit C to assess both inhibition of growth cone collapse and its reversal in neurons.

9. This assay identified as the growth cone assay is based on the common effect on neurons of three widely prescribed drugs (lithium, valproic acid, and carbamazepine) used for the treatment of bipolar disorder. More specifically, as described on Page 5 of the article in Exhibit C, the action of the test drugs which exhibit a positive response, such as the aforementioned anti-convulsants "on growth cones parallels their clinical efficacy in the treatment of bipolar disorders..." As shown by these experiments, lithium, valproic acid and carbamzepine each increase the spread area of growth cones of neurons derived from the rat cerebral cortex which effects are reversed by inositol. Furthermore, this assay is consistent with the finding that gabapentin and phenytoin, two other anti-convulsant drugs, which are not commonly used to

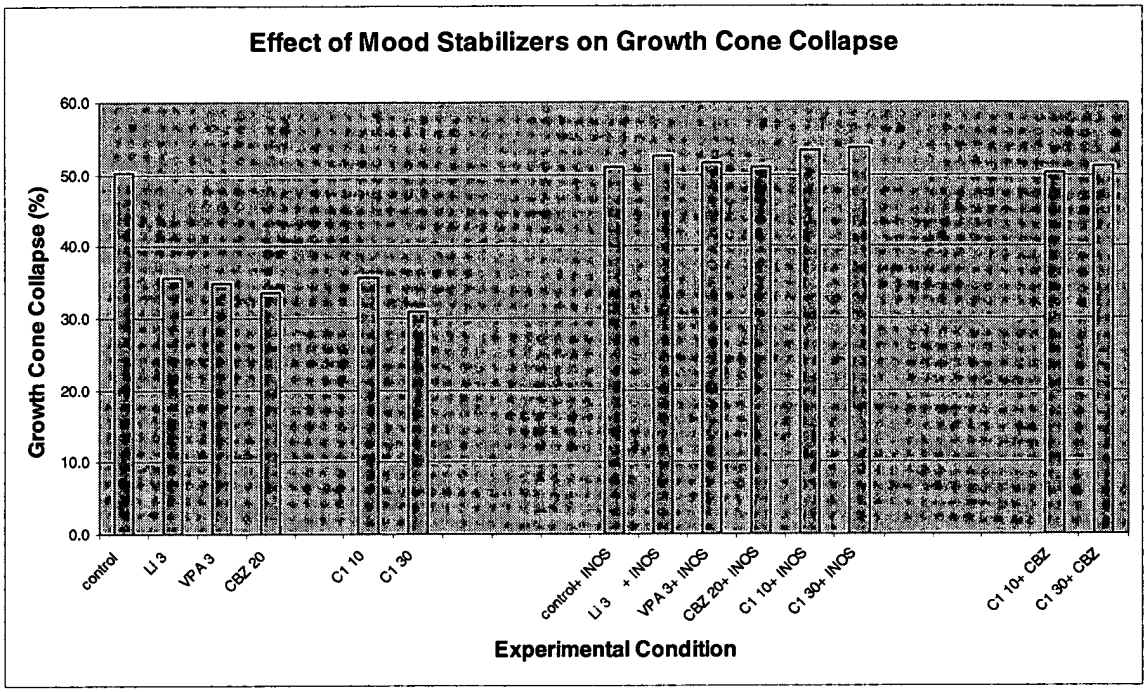
treat bipolar disorder, do not mimic the effects of the mood stabilizers, e.g., valproic acid, lithium or carbamazepine, on sensory neuron growth cones.

10. Thus, the assay can differentiate between those drugs which mimic the effects of the mood stabilizers on sensory neuron growth cones and those which do not so mimic. Those that mimic the effect of mood stabilizers on sensory neuron growth cores are indicated to be useful for treating bipolar disorder.

11. The procedure of the assay is as follows: Briefly, dorsal root ganglia from newborn rats were cultured as explants plated on PDL- and laminin-coated coverslips in serum-free medium with nerve growth factor. The compounds tested–Compound 1, lithium, valproic acid (VPA), carbamazepine (CBZ), and vehicle control–were added after approximately 24 hours at the doses indicated below. After approximately 18 hours, the sensory neuron axons that extend from the ganglia were loaded with Calcein and then fixed. The number of collapsed growth cones was counted using a Zeiss fluorescence microscope and the growth cone spread area was calculated.

12. Compound 1 was also tested for reversal of the effect of CBZ on growth cone collapse.

13. The results from both experiments are shown below.



(Concentrations for Compound 1 (C1) and CBZ are expressed as μM; concentrations for lithium and VPA are expressed as mM)

14. As shown by the data, Compound 1 behaved in a similar fashion to the known mood stabilizers lithium and VPA. Compound 1, like lithium, VPA, CBZ, and other drugs effective for the treatment of the manic component of bipolar disorder, inhibited the collapse of the growth cones and this inhibition was reversed by increasing extra-cellular inositol.

15. As displayed in the two bars at the far right of the graph, Compound 1 also reversed the effect of CBZ on growth cone collapse; this effect is similar to that observed in this model with compounds (like lithium and VPA) that also have an effect on the depressant component of bipolar disorder.

16. Thus, the experiments demonstrate that Compound 1 behaves in the present assay like lithium and VPA, which have mood stabilizing activity and affect both components of bipolar disorder [bipolar mania and bipolar depression].

17. A second set of experiments utilized a different representative compound described in the instant specification and in Paragraph 4 hereinabove, viz, 2-(acetylamino)-2-

(methoxyamino)-N-benzylacetamide, which is designated as Compound 2 herein. This second set of experiments is an in vivo assay conducted in mice.

18. The method utilized follows the procedure described by Costall *et al.* in *Brain Research*, 123, 89-111 (1977), the contents of which are incorporated by reference. It uses an activity meter similar to that described by Boissier and Simon in *Arch. Int. Pharmacodyn.*, 158, 212-221 (1965).

19. The procedure of the assay is as follows: Male Rj: NMRI mice (10 per group) were injected with d-amphetamine (3mg/kg i.p.) and were placed immediately in an activity meter. The activity meter consisted of 24 covered plexiglass enclosures (25 x 20 x 9 cm), each equipped with two photocell assemblies contained within a darkened enclosure and connected to silent electronic counters. The number of interruptions by each animal (one per cage) of the photo-electric beams was recorded by computer at 10 minute intervals for 30 minutes. The scores were cumulated over the 30 minute period. The test was performed blind.

Compound 2 was evaluated at 10, 20, and 40 mg/kg administered i.p. 30 minutes before amphetamine administration. Lithium carbonate (4 mEq/kg) was administered under the same experimental conditions, and was used as a comparative agent.

20. The results are tabulated hereinbelow:

TABLE 1

TREATMENT (mg/kg) i.p. -30 min	**TREATMENT (mg/kg) i.p.**	**NUMBER OF PHOTO-BEAMS CROSSED (0 to 30 min)**						
		Mean ± s.e.m.			**p value**	**% change from control**		**% antagonism**
Vehicle (saline)	Vehicle	251.7 ± 17.5						
Vehicle	Amphetamine (3)	630.9 ± 102.8	**	(a)	0.002	+151%	(a)	-
Compound 2 (10)	Vehicle	220.6 ± 19.4	NS	(a)	0.249	-12%	(a)	-
Compound 2 (20)	Vehicle	199.2 ± 30.0	NS	(a)	0.148	-21%	(a)	-
Compound 2 (40)	Vehicle	159.8 ± 19.7	**	(a)	0.003	-37%	(a)	-

Compound 2 (10)	Amphetamine (3)	402.8 ± 54.3	NS	(b)	0.065	-36%	(b)	52%
Compound 2 (20)	Amphetamine (3)	428.3 ± 82.3	NS	(b)	0.141	-32%	(b)	40%
Compound 2 (40)	Amphetamine (3)	173.9 ± 31.3	***	(b)	0.000	-72%	(b)	96%
Lithium (4 mEq/kg)	Vehicle	65.1 ± 10.2	***	(a)	0.000	-74%	(a)	-
Lithium (4 mEq/kg)	Amphetamine (3)	241.4 ± 43.1	**	(b)	0.003	-62%	(b)	54%

NS = Not Significant; * = $p < 0.05$; ** = $p < 0.01$; *** = $p < 0.001$

(a): compared with control without amphetamine
(b): compared with control with amphetamine

21. The data show that amphetamine (3mg/kg i.p) induced clear hyperactivity (+151 %). In addition, the data show that Compound 2 (10, 20, and 40 mg/kg i.p.), administered alone, moderately decreased locomotion. Furthermore, Compound 2 (10, 20 and 40 mg/kg i.p.), co-administered with amphetamine, tended to antagonize amphetamine–induced hyperactivity at all doses, with the most marked antagonism (96 %) being observed at 40 mg/kg. Lithium (4 mEq/kg i.p.) also decreased locomotion when administered alone and significantly antagonized amphetamine-induced hyperactivity (54 %).

22. Based on the data, it is clear that the Compound 2 behaved similarly to lithium in this assay.

23. Since Compound 2 behaves like lithium, and since lithium is used to treat bipolar disorder, one can conclude that Compound 2 is useful for treating bipolar disorder.

24. The compounds tested in both assays are representative of the compounds described in Paragraph 4 herein.

25. The data clearly illustrates that compounds of the present invention are pharmacologically active in the growth cone assay and in an animal model assay, both of which responds to lithium, a current treatment for bipolar disorder.

26. This data in both sets of experiments clearly support the allegations in the instant application that compounds described in the instant specification and in Paragraph 4 herein are effectively useful for the treatment of bipolar disorder.

27. The undersigned declares further that all statements made herein of his own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

_____ _____

Dated Robert Harris